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INVESTIGATION OF GAMMA IRRADIATON AS A PLANETARY PROTECTION MICROBIAL REDUCTION PROCESS

PI: Laura Newlin
Co-I: Fei Chen
Zachary Dean
Raffaele Gradini
Charlotte Spry
Kristina Stott

Jet Propulsion Laboratory/California Institute of Technology
Pasadena, CA

JPL

Agenda

- Background Information
  - Planetary Protection (PP)
  - What is Gamma Irradiation?
  - Need for Gamma Irradiation
- Methods
- Results
- Conclusion and Recommended Future Investigations
- Acknowledgements

Background Information: Planetary Protection

What is Planetary Protection (PP)?

- To protect the planets (and science) by preserving them as a target of biological exploration
  - Includes forward (outbound) and backward (Earth return) missions
  - Prevents false positive findings by life detection missions

Meeting the Biological Cleanliness requirements for Mars and Icy satellites:

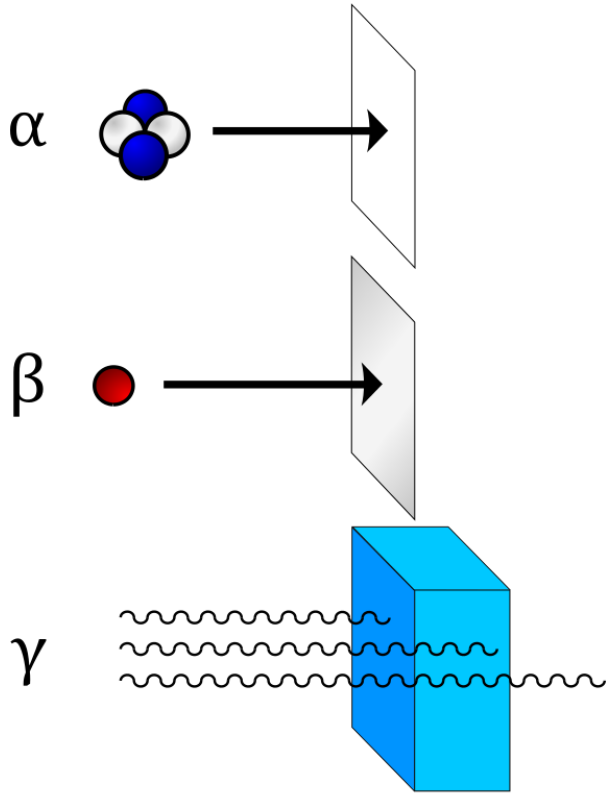
- In order to meet the PP requirements
  - Spacecraft components need to be cleaned / microbially reduced and protected from recontamination
  - Biological cleanliness is a key / driving requirement throughout the entire lifecycle of the project—from Pre-Phase A until spacecraft disposal.

Background Information: Planetary Protection

- Need:
  - Heat Microbial Reduction (HMR) is the primary microbial reduction method
  - Gamma provides penetrating microbial reduction alternative for hardware sensitive to HMR:
    - Antennas
    - Batteries
    - Reaction wheel lubricants
    - Etc.

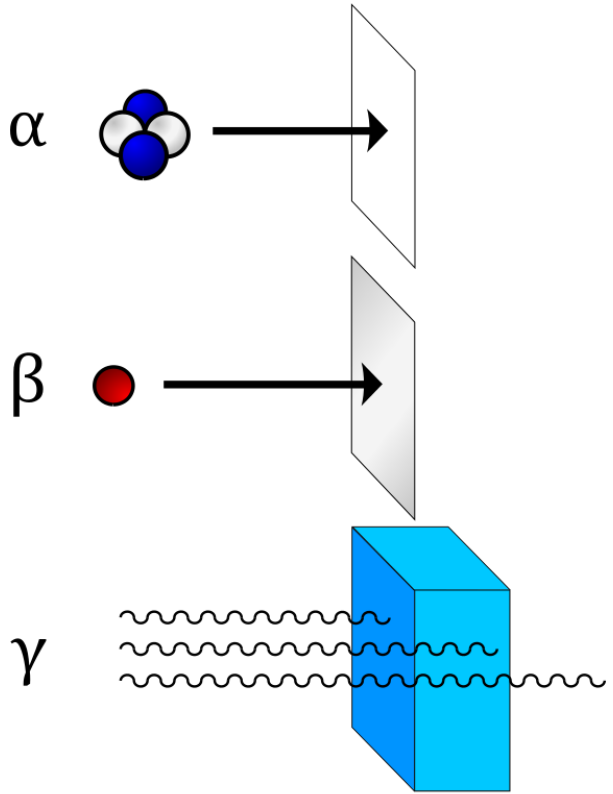
Background Information: Gamma Irradiation

- Alpha particles are larger—2 protons and 2 neutrons (charged helium atom)
- Beta particles are electrons
  - Also break bonds (ionizing)
  - Less penetrative than gamma, more than alpha
- Gamma is photon energy
  - Same as light energy (UV radiation, etc.), but gamma rays carry much more energy
    - Unlike UV, gamma breaks molecular bonds

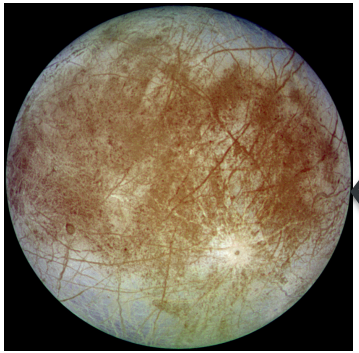


Background Information: Gamma Irradiation

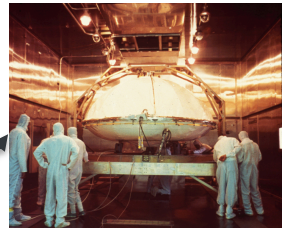
- Typically carry energy > 100keV
- Important:
  - Typical gamma sources do not cause hardware to become radioactive
    - The energy is too low



Background Information: Need



Europa as seen from Galileo



Heat microbial reduction (HMR):

- NASA PP approved protocol
- Can treat bulk and surfaces of hardware

Vapor hydrogen peroxide (VHP):

- NASA PP approved protocol (use less common than HMR)
- Can only treat hardware surfaces

Gamma Irradiation (λ):

- NASA PP protocol not yet established
- Can treat bulk and surfaces of hardware
- Useful for Jovian missions, for hardware not compatible with heat (chemical reactions, etc.), and when more microbial reduction is needed than HMR can provide alone

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Background Information: Need

Advantages	Disadvantages
Low Temperature	Not Validated
Predictable and Repeatable	Cost
No Radiation Byproduct or Residual	Polymers & glasses can be affected*
No Further Process Required	Radioactive source
Volumetric	
Time	

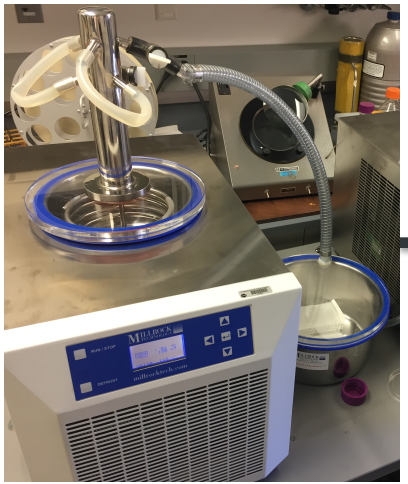
* Polymers which are radiation stable are very expensive

Methods

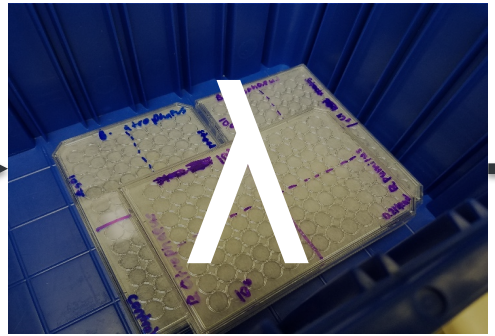
- Selected organisms based on the following criteria:
  - Known radiation resistance
  - Desiccation resistance
  - Heat resistance
  - Available in the JPL PP archive
- Both spore formers and non-spore formers were selected:
  - *Deinococcus radiodurans* (most radiation resistant organism known)
  - *Bacillus pumilus* (gamma indicator organism)
  - *Bacillus atrophaeus,* ATCC 29669 (heat indicator organisms)
  - *Geobacillus stearothermophilus* (VHP indicator organism)
- Overall goal:
  - Determine dose to eliminate extremophiles, assess *D*-values

Methods

- High-level procedure:



Dry organisms on plate



Expose to gamma

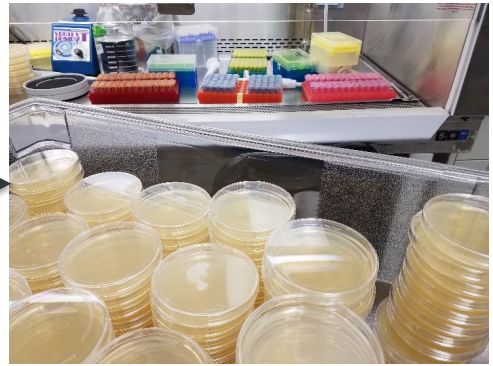
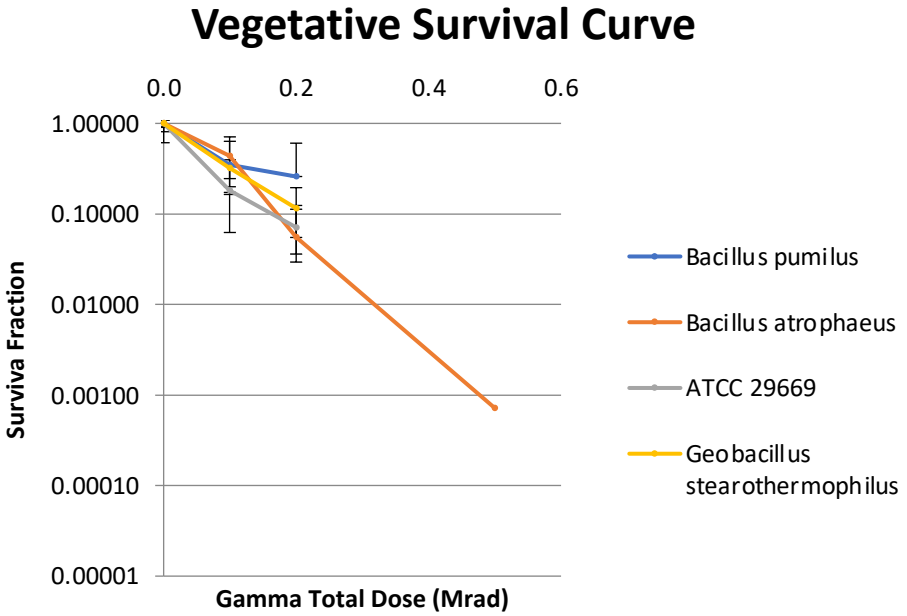
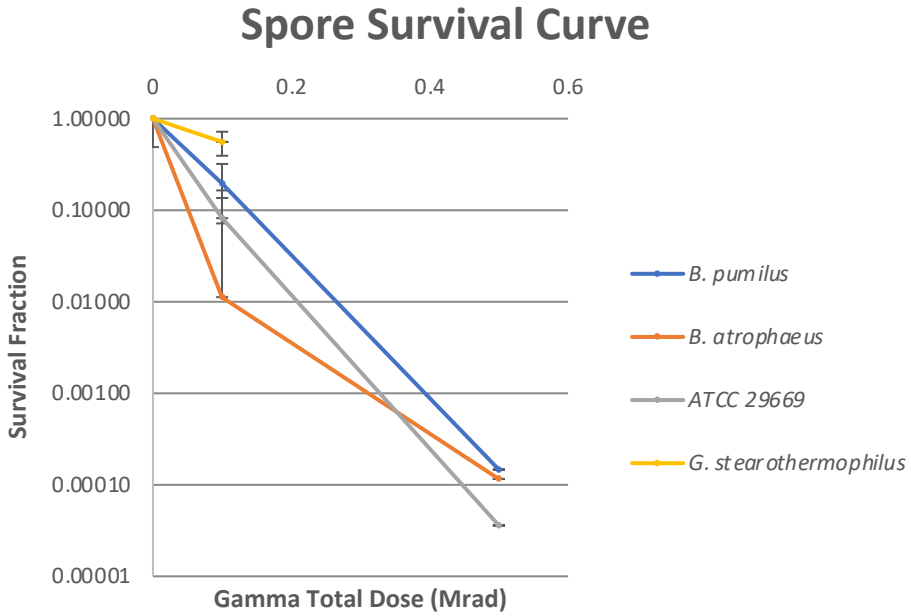


Plate and observe growth

Results

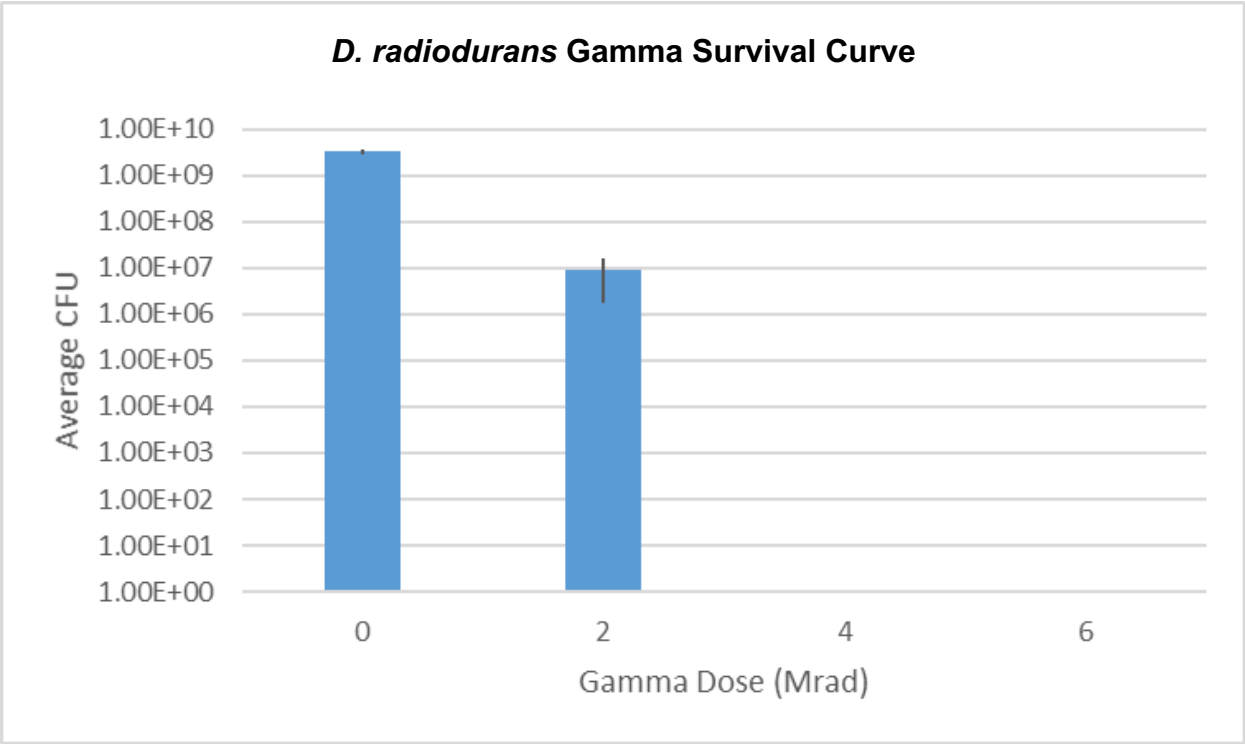
- Preliminary results (curves end where no growth occurred)





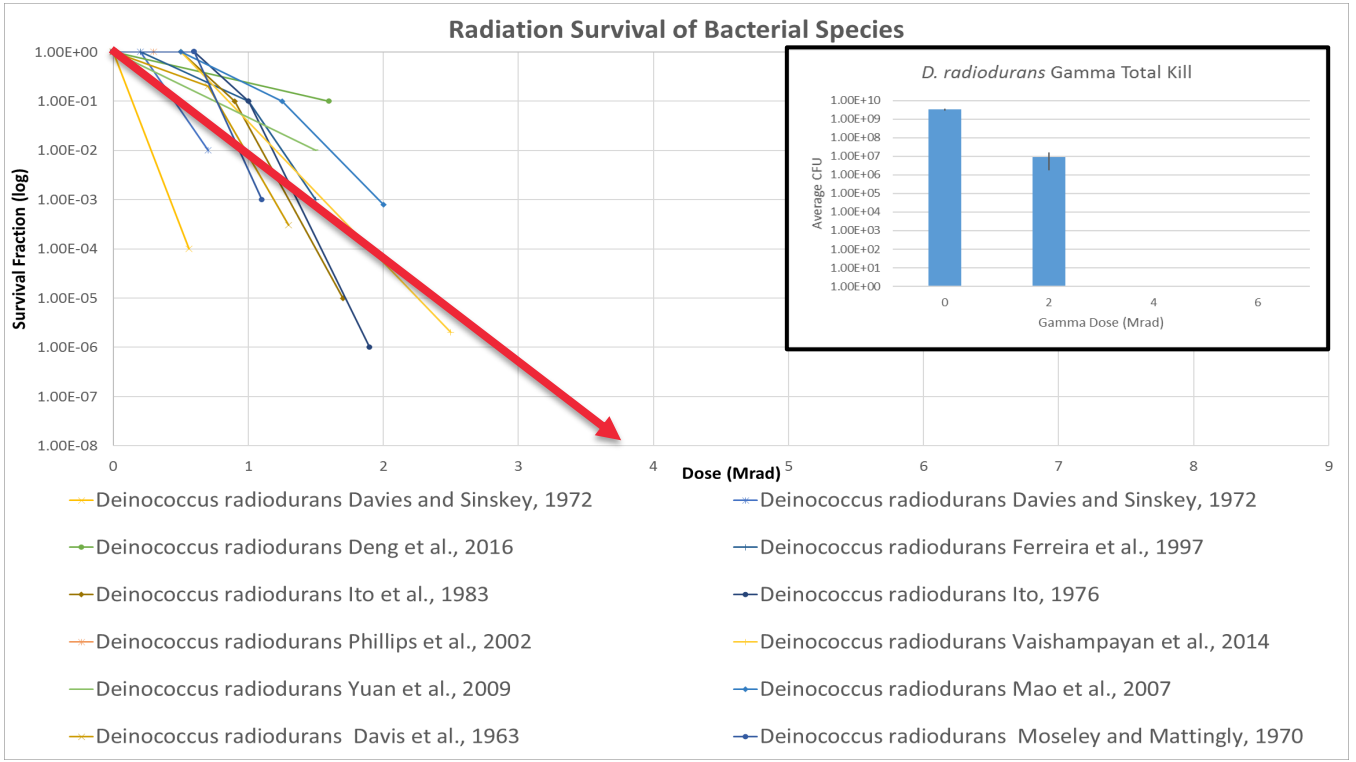
Results

- Preliminary results (no growth at 4, 6 Mrad)
  - *D. radiodurans* is the most radiation resistant organism known



Results

Literature Study on Results (courtesy Reuhle, M. & Park, H.):



Conclusion and Recommended Future Investigations

Conclusion

- Gamma Irradiation is a viable protocol that is useful for microbial reduction
  - Particularly in situations where HMR is not possible

Recommended Future Investigations

- Materials compatibility
  - Only requires a dosimeter (no organisms)
  - Effective measurement of maximum dose that hardware can take
- Determine exposure needed for reduction in bulk materials
- Establish NASA PP approved protocol for gamma irradiation as a sterilization modality

Acknowledgements

- The research was carried out at the Jet Propulsion Laboratory, California Institute of Technology
- The JPL team contributing to this study were: Zach Dean, Laura Newlin, Kristina Stott, Fei Chen, Raffaele Gradini, and Charlotte Spry.

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California Institute of Technology

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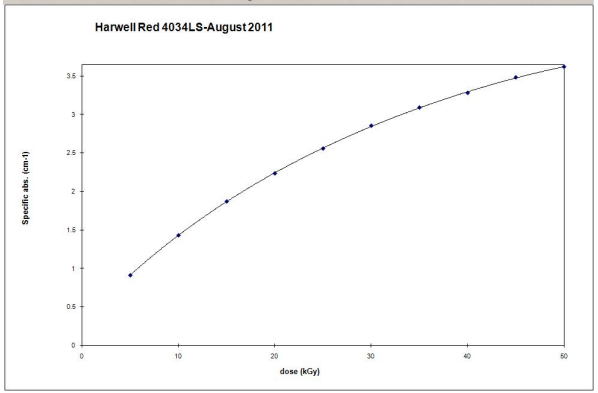
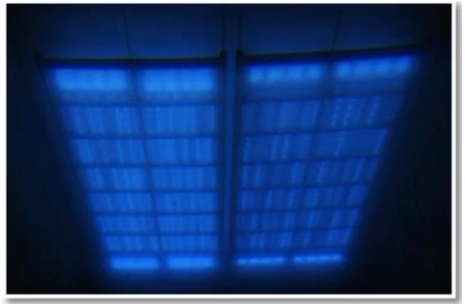
Back-up Slides

Radiation Dosimeters (cont'd)

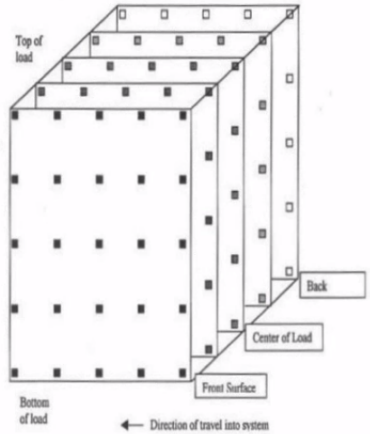


- Used for for chamber and dose-validation
- Inexpensive: $1236 per 1000
- 0.5 - 5 Mrad dose range (matches our required range)
- Easy to use:
  - Spectrometer measurement
  - Small, can be attached to H/W easily
- Quality-controlled:
  - Each batch is calibrated (and shipped with calibration curve) at the National Physical Laboratory (Teddington, England)
    - Curve (bottom-left)
- Used by Steris

Radiation Dosimeters (cont'd)



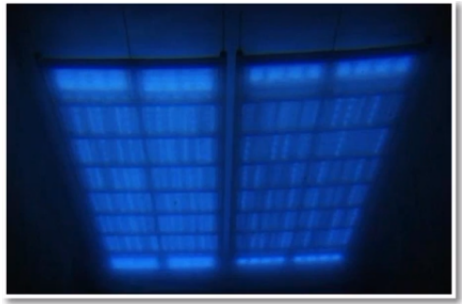
Dose distribution within a Co-60 irradiator (each is unique)



Dosimeters placed in 3D configuration on hardware

- Positioned around hardware to determine actual maximum dose received during sterilization
  - To compare this number to the sterilization dose required
- Uniformity of dose distribution (dose-mapping) on hardware also measured
  - Dose Uniformity Ratio = Max Measured Dose / Min Measured Dose
    - Always > 1, but should not increase over 2
  - More 3-D resolution needed for more complex hardware

Radiation Dosimeters (cont'd)



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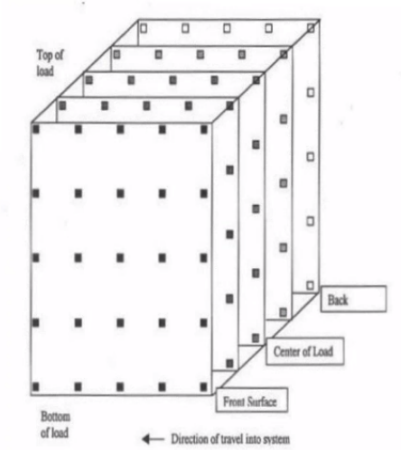
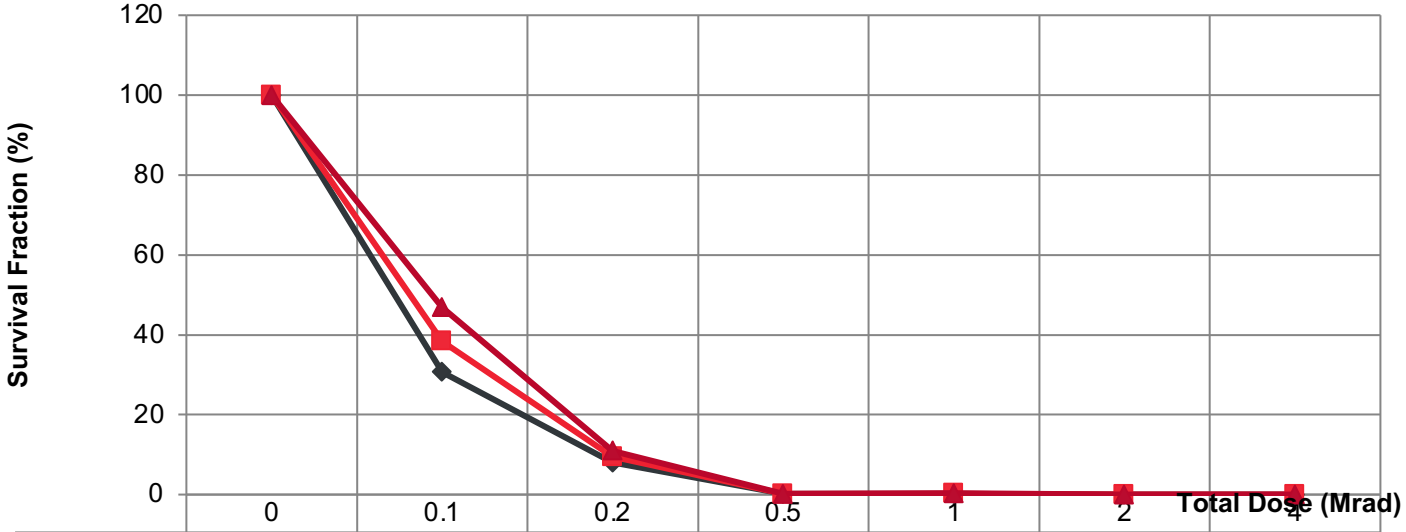


Dosimeters placed in 3D configuration on hardware

- Reference dose-mapping can be done with surrogate H/W (reference hardware)
  - Done to simulate product placement in packaging (bagging, etc.)
  - Prevents recontamination retrieving dosimeters after H/W exposure in gamma
  - However, must be in triplicate
- New mapping should be done when irradiator rack is changed
- Plan for gamma materials testing this summer will use surrogate H/W
  - No organisms

Down-select Experiment (cont'd)

D-Value – *Deinococcus radiodurans*



	0	0.1	0.2	0.5	1	2	4
Best	100	30.68	8.04	0.12	0.13	0	0
Average	100	38.36	9.42	0.12	0.22	0	0
Worst	100	46.83	10.95	0.13	0.31	0	0

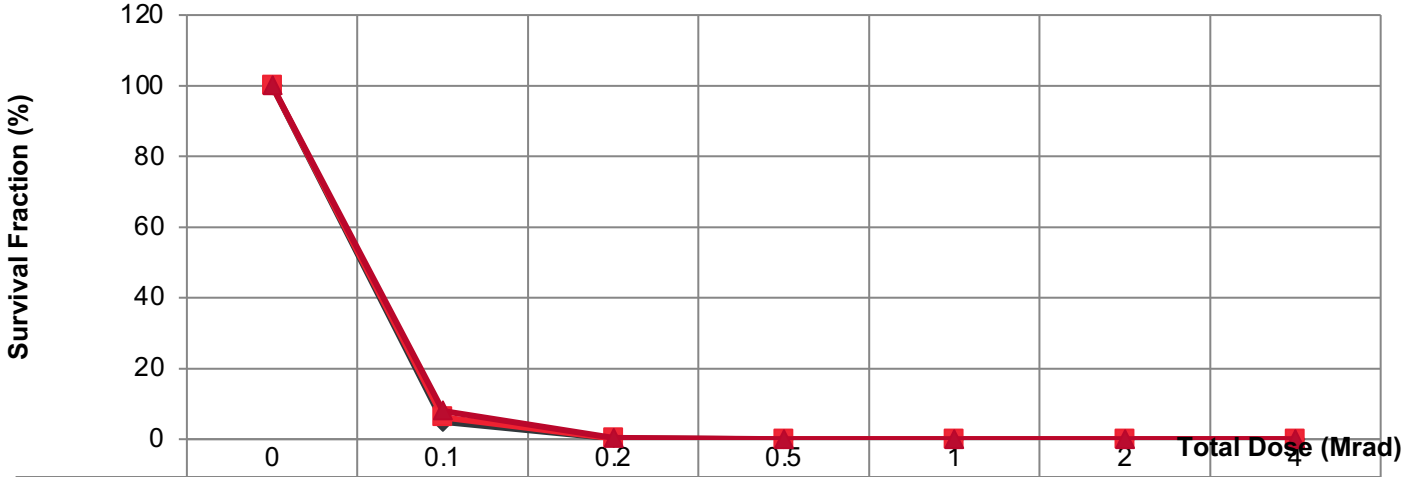
Down-select Experiment (cont'd)

D-Value – *Acinetobacter radioresistens* WC-A-157

	0	0.1	0.2	0.5	1	2	4
Best	100	10.20	0.73	0.00	0.00	0	0
Average	100	11.70	0.86	0.00	0.00	0	0
Worst	100	13.21	0.99	0.00	0.00	0	0

Down-select Experiment (cont'd)

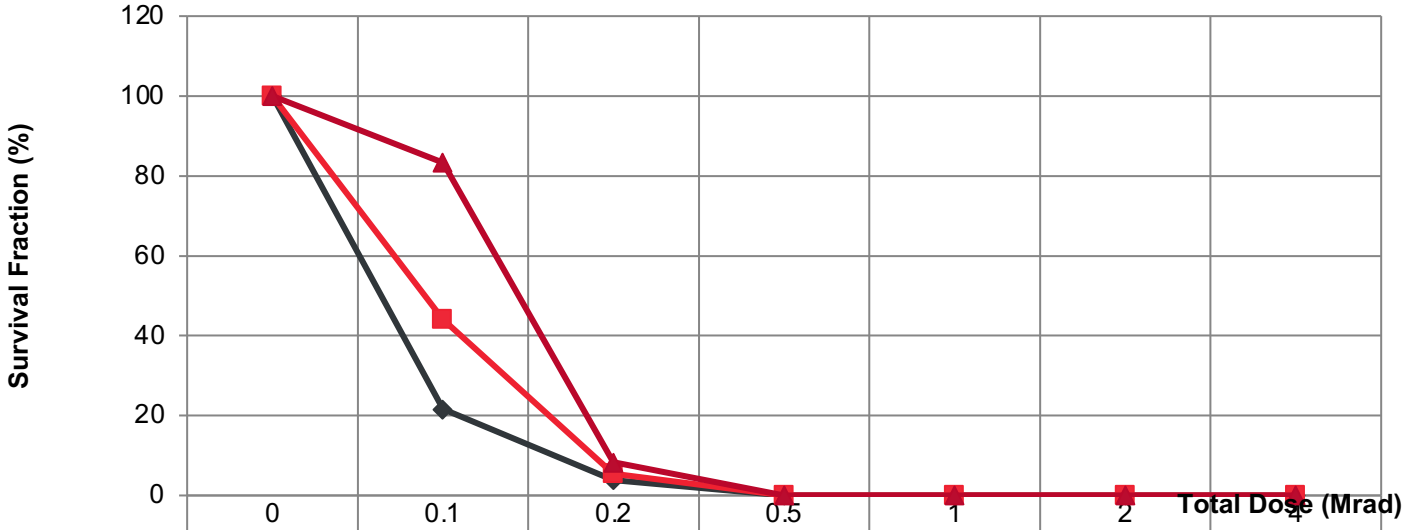
D-Value – *Staphylococcus epidermidis strain* F71028



	0	0.1	0.2	0.5	1	2	4
Best	100	4.82	0.24	0.00	0.00	0	0
Average	100	6.18	0.28	0.00	0.00	0	0
Worst	100	8.05	0.33	0.00	0.00	0	0

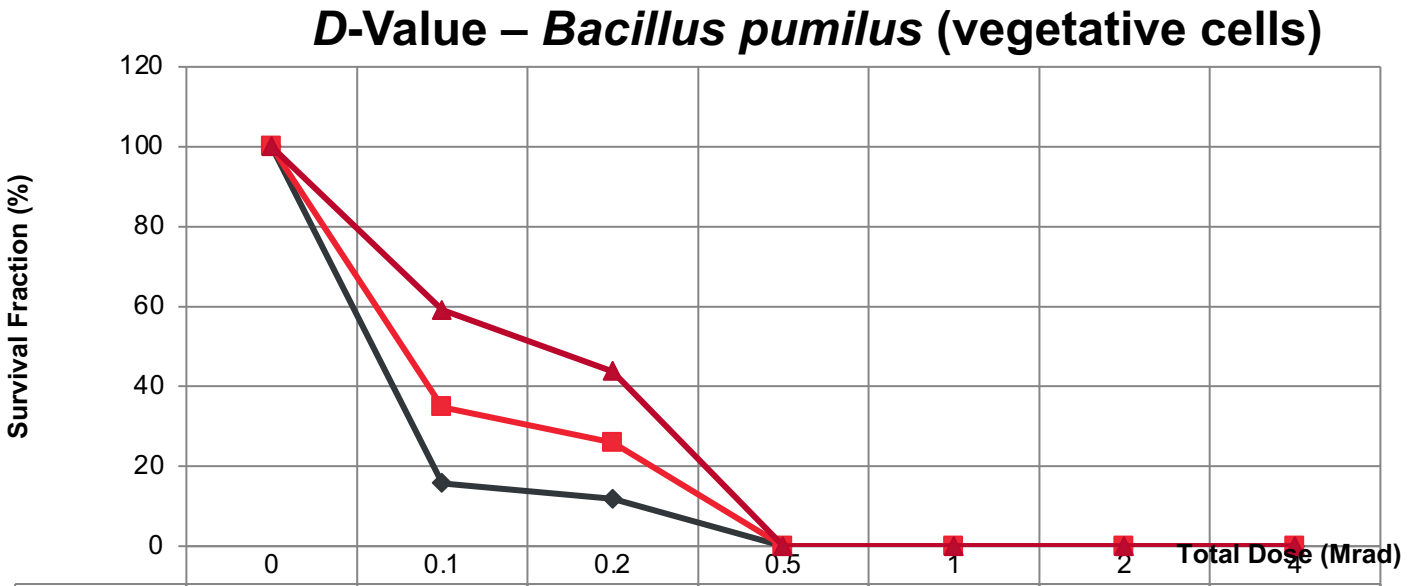
Down-select Experiment (cont'd)

D-Value – *Bacillus atrophaeus* (vegetative cells)



	0	0.1	0.2	0.5	1	2	4
Best	100	21.56	3.91	0.00	0.00	0	0
Average	100	44.12	5.50	0.00	0.00	0	0
Worst	100	83.35	8.27	0.00	0.00	0	0

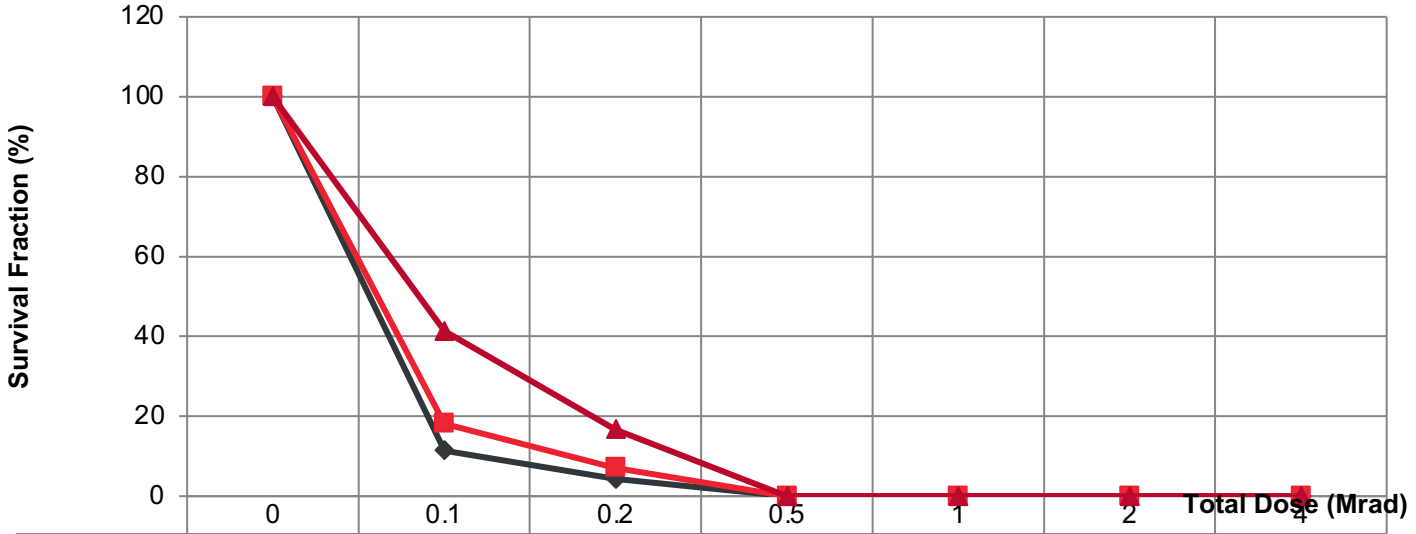
Down-select Experiment (cont'd)



	0	0.1	0.2	0.5	1	2	4
Best	100	15.72	11.83	0.00	0.00	0	0
Average	100	34.83	25.90	0.00	0.00	0	0
Worst	100	59.07	43.75	0.00	0.00	0	0

Down-select Experiment (cont'd)

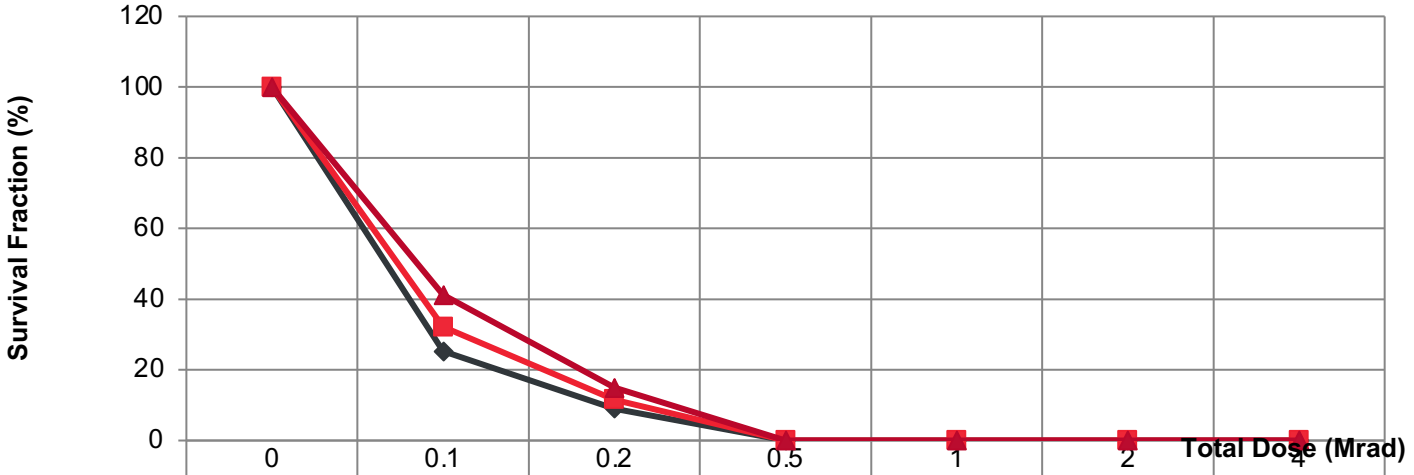
D-Value – ATCC 29669 (vegetative cells)



	0	0.1	0.2	0.5	1	2	4
Best	100	11.44	4.32	0.00	0.00	0	0
Average	100	18.19	7.11	0.00	0.00	0	0
Worst	100	41.35	16.69	0.00	0.00	0	0

Down-select Experiment (cont'd)

D-Value – *Geobacillus stearothermophilus* (vegetative cells)



	0	0.1	0.2	0.5	1	2	4
Best	100	25.22	9.03	0.00	0.00	0	0
Average	100	32.10	11.56	0.00	0.00	0	0
Worst	100	41.15	14.90	0.00	0.00	0	0